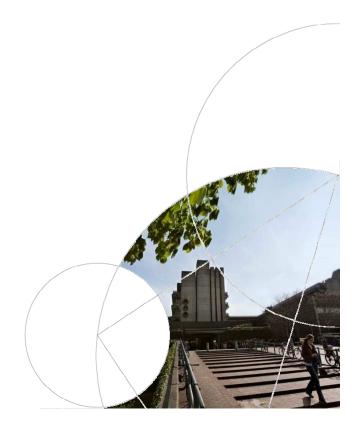


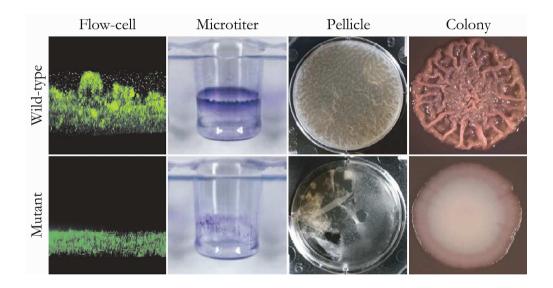
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High throughput biofilm assay

Martin Nilsson Department of International Health, Immunology and Microbiology



Techniques for studying laboratory biofilms





Crystal violet reference

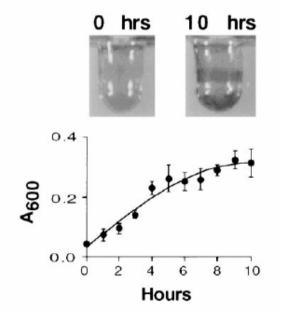
Molecular Microbiology (1998) 28(3), 449-461

Initiation of biofilm formation in *Pseudomonas* fluorescens WCS365 proceeds via multiple, convergent signalling pathways: a genetic analysis

George A. O'Toole and Roberto Kolter*

Department of Microbiology and Molecular Genetics,

Harvard Medical School, 200 Longwood Avenue, Boston,
MA 02115, USA.





Crystal violet biofilm assay

- Bacteria are inoculated in a microtitre dish
- The wells are rinsed after incubation.
- Biofilm cells are stained with a solution of Crystal violet.
- Washing procedure
- Quantified by solubilizing cv in ethanol and determining the absorbance by measuring at 600 nm



Avantages and disadvantages with CV-assay



- easy and cheap to set up
- quntitatively
- large scale possibilities

- does not reflect environmental conditions
 - risk of washing loosely attached cells away
 - lack of possibilites for biofilm structure studies





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High through put equipment



Biomek 2000 (Beckman culter)



Freedom EVO® (Tecan)



Qpix 2 (Genetix)



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Some issues - Shall I go high through put

Amount of clones to analys

Frequently analyses

Identify weak points in the flow system

Obtain matched high trough put – using several systems



